I. AMENDMENTS

In the claims:

19. (Twice Amended) A method for immunizing a vertebrate subject comprising parenterally administering to the vertebrate subject an immunologically effective amount of

a) an adjuvant comprising a detoxified mutant of [a cholera toxin (CT) or] an *E. coli* heat-labile toxin (LT) ADP-ribosylating toxin selected from the group consisting of LT-R72 and LT-K63 in combination with a pharmaceutically acceptable vehicle; and

b) at least one selected antigen.

Please cancel claim 22, without prejudice or disclaimer.

In claim 27, please insert --the-- between "and antigen."

II. REMARKS

Claims 1-19 and 21-30 are presently pending in this application. Claims 1-18 have been withdrawn pursuant to a restriction requirement. Claims 19 and 21-30 stand variously rejected under 35 U.S.C. §§ 112 and 102. Claim 19 has been amended to specify that the detoxified mutant is either LT-R72 or LT-K63. Claim 22 has been canceled, without prejudice or disclaimer. Claim 27 has been amended herein to specify "the" antigen as requested by the Examiner. No new matter is added a result of this amendment and entry thereof is respectfully requested.

Applicants note that these amendments are made without conceding the correctness of the Examiner's position. Applicants reserve the right to pursue the canceled subject matter in a continuation or divisional application.

Overview of the Invention

The pending claims are drawn to methods of immunizing a vertebrate subject by administering an adjuvant comprising a detoxified LT-R72 or LT-K63 mutant of a an *E. coil* heat labile toxin in combination with at least one antigen. Detoxified includes both "completely nontoxic and low residual toxic mutants of the toxin in question. Preferably, the detoxified protein retains a toxicity of less than 0.01% of the naturally occurring toxin courterpart ..." (page 7, lines 26-30).

Another aspect of the present invention is that the adjuvant and antigen are administered parenterally. In other words, as defined by Applicants on page 7, lines 21-26 of the specification, parenteral administration refers to "introduction into the body outside of the digestive tract, such as by subcutaneous, intramuscular, transcutaneous, intradermal, or intravenous administration. This is to be contrasted with adjuvants that are delivered to a mucosal surface, such as oral, intranasal, vaginal, or rectal." Thus, the present invention provides methods using detoxified LT toxins as parenteral adjuvants.

1449 Forms

Applicants have not received an initialed Form 1449 from the IDSs submitted on (1) May 7, 1998; (2) September 11, 1998; and (3) July 18, 2000. Copies of these IDSs are submitted herewith and the Examiner is asked to initial and return the forms.

Rejections Under 35 U.S.C. § 112, First Paragraph

Claims 19, 21, 22 and 24-30 stand rejected under 35 U.S.C. § 112, first paragraph as allegedly containing subject matter which was not described in the specification in such a way as to enable one skilled in the art to use the claimed invention. In particular, it is alleged that it would require undue experimentation to practice the invention using detoxified mutant proteins (see, paragraph 17 of the Office Action). Furthermore, it is alleged that the claims are not enabled throughout their scope (see, paragraph 21 of the

Office Action). Additional enablement rejections related to CT-S109 have been obviated by the amendments herein.

Applicants traverse the relevant remaining rejections and address them in turn below.

Enablement of LT-R72

The Examiner continues to base this rejection on alleged unpredictability in the art, with regard to the toxicity of claimed mutants. Thus, with respect to the LT-R72 mutant, the Examiner contends the certain references teach that this mutant is not detoxified and, in addition, that Applicants own Examples do not provide any data reflective of the detoxified nature of LT-R72. The Examiner also rejects the evidence submitted with the previous response (*i.e.* WO 98/18928) regarding the nature of this mutant.

Applicants submit that Fig 4 of the previously-submitted Exhibit A (WO 98/18928) clearly demonstrates that LT-R72 is a "detoxified" mutant, as measured in a YI cell assay. As noted above, Applicants have defined "detoxified" to include mutants that are "less toxic" than their wild type counterparts, preferably 0.01% of wild-type toxicity as measured, for example, in a Y1 cell assay. Fig 4 of WO 98/18928 plainly shows that these mutants are detoxified -- they 10^5 fold less toxic than wild type as determined in Y1 cell assays. Therefore, Applicants are at a loss as to how the Examiner can state that Figure 4 of this publication "clearly show[s] that LT-R72 mutant is **not** detoxified." (Office Action, page 5, emphasis in original).

Furthermore, the burden is on the Examiner to provide evidence as to why a skilled artisan could not make or use the claimed invention based on the guidance of the specification. Affidavits by experts can be used to establish what the specification reasonably conveys to the skilled artisan. *In re Alton,* 37 USPQ2d 1578, 1584 (Fed. Cir. 1996). As detailed in the attached Declaration by Dr. del Giudice:

5. I believe that one working in this field would clearly consider both LT-R72 and LT-K63 to be "detoxified" mutants. As noted above in paragraph 4, the application defines "detoxified" as mutants having less than 0.01% toxicity of the naturally occurring toxin, for example as measured by the art-recognized YI assay. Example 1 indicates that LT-K63 is inactive "*in vitro* on YI cells." (see, page 28, lines 14-15 of the application). Similarly, LT-R72 is also detoxified, as evidenced by Figure 4 in WO 98/18928 which shows that LT-R72 has much less than 0.01% the toxicity of the naturally occurring toxin. Therefore, both LT-R72 and LT-K63 are detoxified mutants. (del Giudice Declaration, paragraph 5).

In sum, because LT-R72 and LT-K63 are detoxified mutants, this rejection is improper and Applicants respectfully request that it be withdrawn.

The Specification Enables Parenteral Administration of LT Detoxified Mutants

The Office further rejects previous claims 19, 21, 25, 26 and 30 as allegedly not enabled throughout their scope. In particular, it is alleged that the specification only enables intramuscular and subcutaneous administration of LT-K63 and only transcutaneous administration of LT-R72.

Because the specification fully enables parenteral administration of these detoxified mutants, Applicants traverse the rejection and supporting remarks.

A specification is enabling if it teaches one of skill in the art how to make and use the claimed invention without requiring undue experimentation. *In re Wands*, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988). Furthermore, one does not look to the claims, but to the specification to find out how to practice the claimed invention. *W.L. Gore & Assoc., Inc. v. Garlock, Inc.* 220 USPQ 303 (Fed. Cir. 1983). Proof of enablement is required only where adequate reasons are advanced by the examiner to establish that a person skilled in the art could not use the genus as a whole. In addition, the scope of enablement must bear only a reasonable (not exact) correlation to the scope of the claims. *See, e.g., In re Fisher,* 166 USPQ 18 (CCPA 1970) and M.P.E.P. 2164.08. Simply put, it is not required

that Applicants exemplify each and every possible parenteral administration mode for each and every detoxified LT-A mutant in order to satisfy the enablement requirement.

In light of Applicants' specification and the state of the art, a skilled artisan could readily use LT-K63 or LT-R72 as parenteral adjuvants using any number of art-recognized modes of parenteral administration. The term "parenteral" is clearly defined in the specification to (1) include any mode of "introduction into the body outside of the digestive tract" (page 7, line 23) and (2) exclude administration to mucosal surfaces (page 7, lines 25-26). As acknowledged by the Office, the application exemplifies at least three successful modes parenteral administration of these detoxified mutants, including transcutaneous, subcutaneous and intramuscular. Further, additional modes of parenteral administration were well-known at the time of filing, as described for example on pages 22 through 24. In view of the cited references and teachings of the specification, it would not require undue experimentation for the skilled artisan to use these known modes of administration. Thus, Applicants have established both a reasonable correlation between the exemplified methods and those claimed and, in addition, demonstrated that it would not require undue experimentation for a skilled artisan to practice the invention as claimed.

Further evidence of enablement is found in the attached Declaration where Dr. del Giudice states:

> 6. It is further my opinion that one skilled in the art would understand from the application that the claimed detoxified mutants adjuvants could be administered using any known mode of parenteral administration. Again, as noted above in paragraph 4, the application defines "parenteral administration" as non-mucosal administration outside the digestive tract of the subject. The application discusses how to prepare injectable compositions, topically applied compositions, gel formulations, and cites references providing additional parenteral administration techniques. (see, *e.g.*, pages 22-24 of the application and references cited therein). Furthermore, subcutaneous, transcutaneous and intramuscular routes are specifically demonstrated in the Examples of the application. Thus, I believe that, based on the application and level of skill in the art, one

working in this field would be able to immunize a subject with the claimed detoxified adjuvants using any mode of parenteral administration. (del Giudice Declaration, paragraph 6).

Thus, in view of the guidance (including specific examples) provided by the specification, one skilled in the art could easily practice the claimed methods without undue experimentation. Accordingly, withdrawal of this rejection is respectfully requested.

The Cited Reference Does not Establish Unpredictability

Applicants also disagree with the assertion that "the prior art demonstrates the unpredictability of detoxified mutants." (Office Action, page 4). The single cited by the Examiner (Pizza et al) does no such thing. Unlike the claimed methods, Pizza et al. is not directed to the use of detoxified LT mutants as parenteral adjuvants. Moreover, as described above, the burden is on the Examiner to provide evidence as to why it would require undue experimentation to practice the invention as claimed. Pizza's failure to specifically identify the toxicity of CT-S109 certainly does not rise to the level of establishing that the claimed invention is "unpredictable" or that it would require "undue experimentation" to practice the invention. As noted in the attached Rule 132 Declaration:

> 8. It is further my opinion that Pizza et al. is not relevant to the subject matter claimed in the application. Pizza et al. is concerned with analyzing the structure-activity relationship of LT-A mutants. One working in this field would have no reason to apply this information to use of LT mutants as parenteral adjuvants. In particular, Pizza et al. gives LT-R72 a "++" rating in terms of toxicity on YI cells. This is compared to a "+++" rating for wild type. Although, Pizza does not provide absolute values, it is my opinion that one working in this field would not consider a rating of "++" to be "detoxified" as defined in the application. Accordingly, I do not believe that Pizza is relevant to the claimed invention. (del Giudice Declaration, paragraph 8)

Since Pizza is restricted to mucosal administration, and there is no evidence in this reference that establishes unpredictability of the claimed invention, the reliance on this reference is misplaced.

Finally, in light of the evidence and affidavits of record, Applicants request, pursuant to 37 C.F.R. § 1.104(d)(2) that the Office support their rejection and rebut the del Giudice Declaration with specific data and a supporting affidavit.

In view of the foregoing, the rejections under section 112, first paragraph are improper and Applicants respectfully request these rejections be withdrawn.

35 U.S.C. § 112, Second Paragraph

Claims 27 stands rejected under 35 U.S.C. § 112, second paragraph as allegedly indefinite for certain typographical errors and for alleged lack of antecedent basis.

The claim has been amended to obviate this rejection. The amendment has been made without conceding the correctness of the Examiner's position and solely to advance prosecution. In view of the foregoing amendments and remarks, the rejection under section 112, second paragraph has been addressed and Applicants respectfully request it be withdrawn.

Rejections Under 35 U.S.C. § 102

Claims 19-23, and 25-30 are rejected under 102(a) as allegedly anticipated by WO 97/02348, published 01/23/95. WO '348 is cited as allegedly disclosing a method of immunizing a vertebrate subject by administering an effective amount or dose of a composition comprising a detoxified mutant protein of a bacterial toxin.

Applicants traverse. It is well-settled that in order to constitute an anticipatory reference, the cited document must contain an enabling disclosure. *Chester v. Miller*, 15 USPQ2d 1333, 1336 n.2 (Fed. Cir. 1990); see also, *Scripps Clinic & Research Foundation v. Genentech, Inc.*, 18 USPQ2d 1001, 1011 (Fed. Cir. 1991). In other words,

the reference must teach one of skill in the art how to practice the claimed invention, without undue experimentation.

In the case at hand, the disclosure of WO '348 does not enable the claimed methods. As noted above, the claimed methods are specifically directed to methods of parenteral administration and methods in which the detoxified LT or CT mutant acts as an adjuvant. The reference contains no teaching or guidance in either of these regards. Rather, the reference is directed to methods which use certain detoxified CT or LT mutants as antigens. The two statements in WO '348 (page 15 and page 16) which are relied on by the Office as allegedly suggesting use of the mutants as adjuvants, are unsubstantiated, general statements and do not provide the guidance necessary to lead one of skill in the art to the claimed invention.

Further evidence of the insufficiency of WO '348 is submitted with this response. In the attached Rule 132 Declaration, Dr. del Giudice states:

> 7. It is also my opinion that WO '348 does not describe or demonstrate the methods claimed in the application. First, WO '348 does not disclose methods of immunization using the claimed detoxified mutants as adjuvants. Rather, WO '348 is primarily focused on use of detoxified mutants as antigens (see, for example, claim 3 directed to vaccines comprising the detoxified mutant and an adjuvant and claim 4 directed to vaccines comprising the detoxified mutant and a second immunogenic agent). Adjuvanticity is not tested. Second, parenteral administration of the claimed detoxified mutants is not described or demonstrated by WO '348. The immunogenicity of these mutants is described only in mucosal immunization regimes. Certainly, there is no description or suggestion that the claimed mutants could be used as parenteral adjuvants. Accordingly, I do not believe that WO '348 in any way teaches the use of LT-R72 or LT-K63 as parenteral adjuvants. (del Giudice Declaration, paragraph 7).

Thus, because WO '348 does not describe or demonstrate the claimed methods, none of the pending claims are anticipated by this reference and withdrawal of this rejection is respectfully requested.

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III. CONCLUSION

In view of the foregoing, Applicants submit that the claims are now in condition for allowance and requests early notification to that effect.

Please direct all further communications regarding this application to:

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Respectfully submitted,

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